Patent Attorney's Docket No. 028

NT AND TRADEMARK OFFICE
) ) ) Group Art Unit: 1638
) Group Art Unit: 1638
) Examiner: Anne R. Klubelik
) Confirmation No.: 9928
) ) ) )

## RESPONSE TO RESTRICTION REQUIREMENT AND AMENDMENT

Assistant Commissioner for Patents Washington, D.C. 20231

Sir:

This Reply and Amendment is in complete response to the Restriction Requirement set forth in the Official Action (Restriction Requirement) mailed on March 7, 2003 (Paper No. 9).

### IN THE SPECIFICATION:

Kindly replace the paragraph beginning at line 17, page 9, with the following:

Figure 1. Comparison of the deduced Arabidopsis thaliana MinD amino acid sequence with known MinD gene products. Arabidopsis thaliana sequence mzf 18.5, GenBank AB009056; Chlorella vulgaris chloroplast minD, Swissprot Acc No. P56346; Synnechocystis PCC6803 MinD locus, Swissprot Acc. No. q55900, GB3024144;



Escherichia coli, GenBank Acc. No. P18197 (SEQ ID NOS: 1-4). The underlined asterisks indicate a putative nucleotide binding region. Alignments were done first using the Basic Local Alignment Search Tool (NCBI) and further aligned manually.

Kindly replace the paragraph beginning at line 12, page 11, with the following:

Figure 8. Alignment of the *Arabidopsis thaliana* MinE protein with the MinE protein from other organisms. Syne: *Synechocystis* sp. (GenBank BAA10661); Guill: *Guillardia theta* (GenBank AAC35620); Chlorel: *Chlorella protothecoides* (GenBank CAB42593); Ecoli: *Escherichia coli* 0157:H7 (GenBank BAB35091); Neiss: *Neisseria meningitidis* Z2491 (GenBank CAB83414); Pseudo: *Pseudomonas aeruginosa* (GenBank AAG06633) (SEQ ID NOS: 5-11). Symbols in the *Arabidopsis MinE1* gene: the downward-pointing arrow indicates a putative chloroplast transit-peptide-processing site, and the inverted black triangle shows the location of the intron.

Kindly replace the paragraph beginning at line 7, page 20, with the following:

In an effort to determine a role for the *MinD* gene, an *Arabidopsis thaliana* putative *MinD* homologue (*AtMinD*) was isolated. Oligonucleotide primers were synthesized based on the sequence for the bacterial *MinD* gene, MZF18.5, on bacterial chromosome 5 [5' Forward: TCTCGAGAATGGCGTCTCTGAGATTGTTC (SEQ ID NO: 12); 3' Reverse: TTCTAGATTTGCCATTTAGCCGCCAAAG (SEQ ID NO: 14)]. The primers were synthesized to include the ATG start site and the TAA stop codon (underlined above) and to include an *XhoI* and *XbaI* restriction endonuclease site at the 5' and 3' ends,

respectively. Total DNA from the *Arabidopsis thaliana* strain, Columbia, was used for amplification in a standard PCR reaction buffer [20mM Tris-HCl (pH 8.0), 2.0 mM MgCl<sub>2</sub>, 0.25 mM of each dNTP, 100 ng of each primer and 2 units of Taq DNA polymerase (Gibco/BRL, Rockville MD)]. The DNA fragment was cloned into pGEM T-(Promega, Madison, WI) and verified by sequencing. The *Xhol-XbaI MinD* gene fragment was excised from pGEM and cloned into the *Sal*I and *Xba*I sites of the pKYLX71 binary vector containing the caMV 35S<sup>2</sup> promoter for constitutive overexpression (Maiti et al., *Proc. Natl. Acad. Sci. USA 90*:6110-6114 (1993)). All *E. coli* manipulations were carried out in the strain TB1. The pKYLX71:*AtMinD* recombinant plasmid was mobilized into *Agrobacterium tumefaciens* C58C1:pGV3850 by tri-parental mating (Schardl et al., *Gene 61*:1-11 (1987)). *Agrobacterium tumefaciens* mediated transformation of tobacco plants was performed using the protocols described in Schardl et al., (1987).

Kindly replace the paragraph beginning at line 9, page 27, with the following:

Oligonucleotide primers were synthesized based on the sequence found on bacterial artificial chromosomes (BACs) F23O10.25 and F10D13.22 on chromosome 1. The *Arabidopsis MinE1* gene was isolated by reverse transcription (RT)-PCR from RNA isolated from leaf tissues of *Arabidopsis thaliana* (L.) Heynh., accession line Columbia. For cloning into the KYLX71:35S² binary vector (Maiti et al., *Proc. Natl. Acad. Sci USA 90*:6110-6114 (1993)), primers (5' forward: 5'-AGT TTC TCG GTA *ATG* GCG ATG T-3' (SEQ ID NO: 14); 3' back: 5'-GAC TGT GCC TTT *TCA* TCA CTC T-3' (SEQ ID NO: 15)) were synthesized to include the ATG start site and TAG stop codon (shown in

Application No. 10/067,989 Attorney's Docket No. 028750-219 Page 4

bold italics) with an addition of an *XhoI* and *XhaI* restriction endonuclease site at the 5' and 3' end, respectively, for the sense primers, and reversed for the antisense primers. For green fluorescent protein (GFP) fusion protein the same 5' primer above was used with a 3' primer (5' - TTG AGC TCA CCT CCA ACA TTA AAA TCG AAC CTG-3'(SEQ ID NO: 16) that deleted the stop codon and contained an *SstI* endonuclease site immediately following to provide an in-frame sequence with the entire GFP protein. The GFP gene was isolated by PCR from a plasmid carrying *mgfp5* (Siemering et al 1996); primers (5' forward: 5'-TTG AGC TCA TGA GTA AAG GAG AAG AAC T-3'(SEQ ID NO: 17) and 3'back: 5'-TTC TAG ATT ATT TGT ATA GTT CAT CCA TG-3'(SEQ ID NO: 18)) were designed to have *SstI* and *XbaI* restriction-endonuclease recognition sites.



#### REMARKS

Applicants hereby elect with traverse the claims of Group II (Claims 1-7, 10-14, and 28-31), which are drawn to a vector comprising a gene encoding a protein with the same function as the *Arabidopsis* MinD protein, a plant whose nuclear genome has been transformed with a gene encoding a protein with the same function as the *Arabidopsis* MinD protein, and a method of producing the plant.

The specification is also amended herein, as requested by the Examiner in the outstanding Office Action (restriction). Specifically, the specification has been amended to reflect the proper sequences identifiers pursuant to the sequence listing filed on May 24, 2002. Thus, no prohibited new matter is introduced by way of this Amendment.

Turning now to the Restriction Requirement, this election is made with traverse. Under M.P.E.P. § 803, a restriction is proper if the subject matter can be restricted into one of two or more claimed inventions, and these inventions are either independent (M.P.E.P. § 806.04) or distinct (M.P.E.P. § 806.05). However, the second element for a restriction requirement to be proper is that if the search and examination of an entire application can be made without serious burden, the examiner *must* examine it on the merits, even though it includes claims to independent and distinct inventions. This is true even when appropriate reasons exist for a restriction requirement. Furthermore, the Office has not set forth an explanation of how a search of the claimed invention would be burdensome.

In the present application, it is believed that there is a close relationship between the subject matter of the claims of Groups I-V, and so there would be no serious burden on the

Application No. 10/067,989 Attorney's Docket No. 028750-219 Page 6

Examiner to examine all of the claims at this time. The claims of Group I and III relate to a vector comprising a gene encoding a protein with the same function as the *Arabidopsis* MinE protein. The claims of Group II, IV and V relate to a vector comprising a gene encoding a protein with the same function as the *Arabidopsis* MinD protein. It is not believed that there would be a serious burden on the Examiner to examine all of the claims together at this time.

At the very least, applicants request a modification of the Restriction Requirement. Applicants respectfully request that the claims of Groups II, IV and V be examined together. The claims of Group II are drawn to a vector comprising a gene encoding a protein with the same function as the Aribidopsis MinD protein. The claims of Group IV and V are drawn to methods of using the of Group II. As such, there is a very close relationship between the subject matter of these two sets of claims. Thus, there would be no serious burden on the Examiner to examiner the claims of Groups II, IV and V together.

In light of the above, withdrawal of the requirement for restriction between Groups I-V is respectfully requested. Alternatively, applicants request a modification of the Restriction Requirement so that the claims of Groups II, IV and V are examined together. Such action is believed to be in order. Further and favorable consideration of all the claims of record on the merits is respectfully requested.

Application No. 10/067,989 Attorney's Docket No. 028750-219 Page 7

In the event that there are any questions relating to this Response to Restriction Requirement, or the application in general, it would be appreciated if the examiner would telephone the undersigned attorney concerning such questions so that prosecution of this application may be expedited.

Respectfully submitted,

BURNS, DOANE, SWECKER & MATHIS, L.L.P.

Bv:

Deborah H. Yellin Registration No. 45,904

P.O. Box 1404 Alexandria, Virginia 22313-1404 (703) 836-6620

Date: April 7, 2003

Application No. 10/067,989 Attorney's Docket No. 028750-219

Page 1

# Attachment to Response to Restriction Requirement dated April 7, 2003 Marked-up Copy

#### Page 9, Paragraph Beginning at Line 17:

Figure 1. Comparison of the deduced *Arabidopsis thaliana* MinD amino acid sequence with known *MinD* gene products. *Arabidopsis thaliana* sequence mzf 18.5, GenBank AB009056; *Chlorella vulgaris* chloroplast minD, Swissprot Acc No. P56346; *Synnechocystis* PCC6803 MinD locus, Swissprot Acc. No. q55900, GB3024144; *Escherichia coli*, GenBank Acc. No. P18197 (SEQ ID NOS. 1-4). The underlined asterisks indicate a putative nucleotide binding region. [The red indicates amino acid identity with the *Arabidopsis* MinD deduced amino acid sequence (in green), and those in blue indicate conserved amino acid substitutions.] Alignments were done first using the Basic Local Alignment Search Tool (NCBI) and further aligned manually.

## Page 11, Paragraph Beginning at Line 12

Figure 8. Alignment of the *Arabidopsis thaliana* MinE protein with the MinE protein from other organisms. Syne: *Synechocystis* sp. (GenBank BAA10661); Guill: *Guillardia theta* (GenBank AAC35620); Chlorel: *Chlorella protothecoides* (GenBank CAB42593); Ecoli: *Escherichia coli* 0157:H7 (GenBank BAB35091); Neiss: *Neisseria meningitidis* Z2491 (GenBank CAB83414); Pseudo: *Pseudomonas aeruginosa* (GenBank AAG06633) (SEQ ID NOS: 5-11). Symbols in the *Arabidopsis MinE1* gene: the downward-pointing arrow indicates a putative chloroplast transit-peptide-processing site, and the inverted black triangle shows the location of the intron.

Attorney's Docket No. 028750-219

## Attachment to Response to Restriction Requirement dated April 7, 2003 Marked-up Copy

## Page 20, Paragraph Beginning at Line 20

In an effort to determine a role for the MinD gene, an Arabidopsis thaliana putative MinD homologue (AtMinD) was isolated. Oligonucleotide primers were synthesized based on the sequence for the bacterial MinD gene, MZF18.5, on bacterial chromosome 5 [5' Forward: TCTCGAGAATGGCGTCTCTGAGATTGTTC (SEQ ID NO: 12); 3' Reverse: TTCTAGATTTGCCATTTAGCCGCCAAAG (SEQ ID NO: 14]. The primers were synthesized to include the ATG start site and the TAA stop codon (underlined above) and to include an *XhoI* and *XbaI* restriction endonuclease site at the 5' and 3' ends, respectively. Total DNA from the Arabidopsis thaliana strain, Columbia, was used for amplification in a standard PCR reaction buffer [20mM Tris-HCl (pH 8.0), 2.0 mM MgCl<sub>2</sub>, 0.25 mM of each dNTP, 100 ng of each primer and 2 units of Taq DNA polymerase (Gibco/BRL, Rockville MD)]. The DNA fragment was cloned into pGEM T-(Promega, Madison, WI) and verified by sequencing. The Xhol-Xbal MinD gene fragment was excised from pGEM and cloned into the SalI and XbaI sites of the pKYLX71 binary vector containing the caMV 35S<sup>2</sup> promoter for constitutive overexpression (Maiti et al., Proc. Natl. Acad. Sci. USA 90:6110-6114 (1993)). All E. coli manipulations were carried out in the strain TB1. The pKYLX71:AtMinD recombinant plasmid was mobilized into Agrobacterium tumefaciens C58C1:pGV3850 by tri-parental mating (Schardl et al., Gene 61:1-11 (1987)). Agrobacterium tumefaciens mediated transformation of tobacco plants was performed using the protocols described in Schardl et al., (1987).

## Attachment to Response to Restriction Requirement dated April 7, 2003

## Marked-up Copy

#### Page 27, Paragraph Beginning at Line 9

Oligonucleotide primers were synthesized based on the sequence found on bacterial artificial chromosomes (BACs) F23O10.25 and F10D13.22 on chromosome 1. The Arabidopsis MinE1 gene was isolated by reverse transcription (RT)-PCR from RNA isolated from leaf tissues of Arabidopsis thaliana (L.) Heynh., accession line Columbia. For cloning into the KYLX71:35S<sup>2</sup> binary vector (Maiti et al., *Proc. Natl. Acad. Sci USA* 90:6110-6114 (1993)), primers (5' forward: 5'-AGT TTC TCG GTA ATG GCG ATG T-3' (SEQ ID NO: 14); 3' back: 5'-GAC TGT GCC TTT TCA TCA CTC T-3' (SEQ ID NO: 15)) were synthesized to include the ATG start site and TAG stop codon (shown in bold italics) with an addition of an XhoI and XbaI restriction endonuclease site at the 5' and 3' end, respectively, for the sense primers, and reversed for the antisense primers. For green fluorescent protein (GFP) fusion protein the same 5' primer above was used with a 3' primer (5' - TTG AGC TCA CCT CCA ACA TTA AAA TCG AAC CTG-3' (SEQ ID NO: 16) that deleted the stop codon and contained an SstI endonuclease site immediately following to provide an in-frame sequence with the entire GFP protein. The GFP gene was isolated by PCR from a plasmid carrying mgfp5 (Siemering et al 1996); primers (5' forward: 5'-TTG AGC TCA TGA GTA AAG GAG AAG AAC T-3' (SEQ ID NO: 17) and 3'back: 5'-TTC TAG ATT ATT TGT ATA GTT CAT CCA TG-3' (SEO ID NO: 18)) were designed to have SstI and XbaI restriction-endonuclease recognition sites.

APR 0 7 2003

Attorney's Docket No. 028750-219

## IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re P	atent Application of	NON-FEE AMENDMENT			
Randy	DINKINS et al.	) Group Art Unit: 1638			
Applic	ation No.: 10/067,989	) Examiner: Anne R. Klubelik			
	February 8, 2002	Confirmation No.: 9928			
For:	TRANSGENIC PLANTS EXPRESSING MinD OR MinE AND AN EFFICIENT METHOD FOR PLANT CHLOROPLAST TRANSFORMATION AND GENE EXPRESSION	) ) )			
	AMENDMENT/REPLY TR	ANSMITTAL LETTER			
	nt Commissioner for Patents ngton, D.C. 20231				
Sir:					
Er	nclosed is a reply for the above-identified pate	ent application.			
[	] A Petition for Extension of Time is also e	enclosed.			
[	A Terminal Disclaimer and the [ ] \$55.00 C.F.R. § 1.20(d) are also enclosed.	(2814) [ ] \$110.00 (1814) fee due under 37			
[	] Also enclosed is/are	<del></del>			
[	Small entity status is hereby claimed.				
[	Applicant(s) request continued examination [ ] \$375.00 (2801) [ ] \$750.00 (1801) fee	on under 37 C.F.R. § 1.114 and enclose the e due under 37 C.F.R. § 1.17(e).			
	[ ] Applicant(s) previously submitted requested.	_, on, for which continued examination is			
[	Applicant(s) request suspension of action exceed three months from the filing of the § 1.103(c). The required fee under 37 C				
]	A Request for Entry and Consideration of (1809/2809) is also enclosed.	f Submission under 37 C.F.R. § 1.129(a)			

[X] No additional claim fee is required.

[ ] An additional claim fee is required, and is calculated as shown below:

AMENDED CLAIMS						
	No. Of CLAIMS	HIGHEST NO. OF CLAIMS PREVIOUSLY PAID FOR	EXTRA CLAIMS	RATE	ADDT'L FEE	
Total Claims		MINUS =		× \$18.00 (1202) =		
Independent Claims		MINUS =		× \$84.00 (1201) =	····	
If Amendment adds multiple dependent claims, add \$280.00 (1203)						
Fotal Amendment Fee						
If small entity status is claimed, subtract 50% of Total Amendment Fee						
TOTAL ADDITIONAL FEE DUE FOR THIS AMENDMENT						

[ ] Charge \$	_to Deposit Account No. 02-4800.
The Commissioner is he	reby authorized to charge any appropriate fees under 37 C.F.R.

[ ] A claim fee in the amount of \$\_\_\_\_\_ is enclosed.

§§ 1.16, 1.17, 1.20(d) and 1.21 that may be required by this paper, and to credit any overpayment, to Deposit Account No. 02-4800. This paper is submitted in duplicate.

Respectfully submitted,

BURNS, DOANE, SWECKER & MATHIS, L.L.P.

y:\_\_\_<u>'</u>

Deborah H. Yellin

Registration No. 45,904

P.O. Box 1404 Alexandria, Virginia 22313-1404 (703) 836-6620

Date: April 7, 2003